# Genotyping of *Campylobacter* isolates from swine, poultry and humans in Canada.

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**Summary** : Direct epidemiological evidence indicating that pork meat can be a source of campylobacteriosis in human are missing, while it is widely accepted for poultry products. A total of 101 *Campylobacter* strains isolated from slaughtered swines, 195 strains isolated from broilers as well as 24 human *Campylobacter* strains from ill patients were included in this study. The genetic characterization of isolates was performed by pulse-field gel electrophoresis (PFGE). Macrorestrictions profiles were obtained with by digestion of DNA with *Kpn*I. The combination of the 72 PFGE swine patterns with the PFGE human patterns gave no genetic link between strains while 20 % of human isolates were genetically related to poultry isolates. Since *C. jejuni* was mainly recovered from human and poultry while swine harbored *C. coli*, the role of swine in the transmission of *Campylobacter* to human population by meat consumption remain uncertain.

**Keywords**: Foodborne pathogen, genetic characterization, swine, poultry, PFGE.

**Introduction**: *Campylobacter* is common bacterial species associated with diarrhea in humans (Aarestrup et al., 1997). In Canada, the rate of campylobacteriosis for 1997 was 44,7 cases per 100 000 inhabitant (Health Canada). It is widely assumed that campylobacteriosis is primarily a food-borne disease. Transmission occurs most commonly via the meat products as a result of fecal contamination during slaughter process (Ketley, 1997). Pigs and broilers are frequently colonized by *Campylobacter* spp. However, their respective contribution to the sporadic human infection by Campylobacteriosis is currently not fully understood (Wassenaar, 1997). To acquire a better knowledge on the origin of campylobacteriosis associated with sporadic cases in human, it is necessary to further characterize the microorganisms involved. Among the available techniques, pulse-field gel electrophoresis (PFGE) is one of the most discriminatory technique for *Campylobacter*. The aim of this study was to genetically and phenotypically characterize the strains recovered from sporadic human clinical cases and healthy swine and broilers within a limited geographical area, during the same period in

order to compare their profiles in order to assess the importance of swine as a source of transmission of *Campylobacter* for humans.

**Material & Methods**: In this study, 101 swine campylobacter isolates and 195 poultry isolates as well as 24 campylobacter from sporadic human clinical cases were studied. PFGE was performed as described previously with some modifications (Gibson et al.,1994). For lysis, the plugs were placed in 2 ml of a solution of lysis buffer with 0,5 mg/ml of lysozyme overnight at 37 °C before digestion by proteinase K (Harrington et al., 1999). Before DNA digestion, a pre-migration was performed by submitting plugs to an electric field of 60 V for 45 min (Whatling & Thomas,1993). Plugs were digested 18 h at 37 °C with 20 U of *Kpn*I restriction endonuclease as recommended by the manufacturer (On et al., 1998). PFGE was performed at 200 V and 14 °C with a pulse time of 5 s for 9 h, 20 s for 9 h and 5 s for 2 h for a total time of 20 h in a Gene Navigator® apparatus. A lambda ladder was used as molecular size marker and reference strains of *C. coli* and *C. jejuni* were also included. DNA bands were visualized under UV light after ethidium bromide coloration and photographed. The pictures were then scanned and analyzed using the Image Master® software version 3.01.

**Results**: All swine isolates were identified as *C. coli*. Among human strains one was identified as *C. coli* while the others were *C. jejuni*. In poultry, *C. jejuni* was mainly recovered. All *Campylobacter* isolates of human origin were analyzed by PFGE and compared with swine and poultry PFGE profiles. When placed in a dendrogram, human profiles were distributed through the 72 different swine genetic profiles with no significant association (under 85 %) or grouped in a small distinct clusters. Only two *C. jejuni* human profiles shared homology at 85 % or more. For comparison of human and poultry *Campylobacter*, macrorestriction profiles revealed that approximately 20 % (5) of human *Campylobacter* isolates were genetically related to genotypes found in poultry.

**Discussion**: Following guidelines of Tenover et al. (1995) for small sets of isolates, a criteria of homology of 85 % was selected to establish relatedness between strains. Strains were considered as closely related if there was two or three different bands between genetic profiles while isolates that differed by one band or less were considered indistinguishable. Since *C. coli* was mostly isolated from swine feces in this study, a low relatedness between swine and human strains was expected. When the swine macrorestriction profiles were compared to human DNA profiles, no relevant association was thus observed. Most of the human isolates were located in separated clusters while some were distributed throughout swine DNA profiles. *C. jejuni* was mostly recovered from human or poultry isolates. In addition, nearly 20% of human isolates were genetically related to poultry isolate. The role of swine in transmission of *Campylobacter* to humans remain uncertain following these

results. On the other hand, the genetic relationship observed between human and poultry isolates indicated that *Campylobacter* isolates from poultry could be linked to human cases of sporadic campylobacteriosis.

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